CONSIDERED: /A.G./

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of:	Confirmation No.: 1312
MARTINEZ *et al.*	Art Unit: 1654
Appl. No.: 10/669,597	Examiner: Gupta, A.
Filed: September 25, 2003	Atty. Docket: 2057.0040002/ELE/GER
For: **Polymer Conjugates with Decreased Antigenicity, Methods of Preparation and Uses Thereof**	

Declaration Under 37 C.F.R. § 1.132 Of Dr. Mark G.P. Saifer

Commissioner for Patents
PO Box 1450
Alexandria, VA 22313-1450

Sir:

I, Mark G.P. Saifer, declare and state as follows:

1. I am named as a co-inventor for the above-captioned U.S. Patent Application No. 10/669,597 ("the '597 application") entitled "Polymer Conjugates with Decreased Antigenicity, Methods of Preparation and Uses Thereof," which has a filing date of September 25, 2003, which claims priority to U.S. Patent Application No. 10/317,092, filed December 12, 2002, and which claims benefit of U.S. Provisional Patent Application No. 60/414,424, filed September 30, 2002.

2. I am currently employed by Mountain View Pharmaceuticals, Inc., the assignee of the above-captioned application, where I hold the position of Vice President and Scientific Director. My credentials are provided in the *curriculum vitae* that is attached to this declaration as Exhibit A. I received my A.B. in Natural Science from the University of Pennsylvania in 1960, and my Ph.D. degree in Biophysics from the University of California at Berkeley in 1966. As seen from my *curriculum vitae*, I have published many papers related to polymer conjugation to bioactive components, including peptides and proteins. Based on my education and experience, I am an expert in the field of polymer conjugation, including PEGylation technologies.

3. I have reviewed and am familiar with the '597 application, the Office Action dated October 4, 2010 ("the Office Action") in the present application, and the currently pending claims, filed in the Reply to Office Action with this Declaration.

4. At page 3 of the Office Action, the Examiner cited Lee *et al.*, U.S. Patent No. 4,261,973 (hereinafter "Lee"). Lee discloses polyethylene glycol ("PEG") at column 4, line 66 to column 5, line 4. The PEG molecules utilized in Examples 1 and 2 of Lee are PEG diols. At column 5, line 2, Lee discloses that PEG was coupled to ovalbumin and to ragweed antigen using cyanuric chloride as the coupling agent.

5. Lee does not disclose compositions in which (a) *at least 95%* of the polyalkylene glycol(s) is (or are) attached to a peptide, protein or glycoprotein at a *single* site on the polyalkylene glycol(s), and (b) a hydroxyl group is present on *at least 95%* of the distal polyalkylene glycol termini in the conjugate. For example, Lee does not disclose capping one end of the PEG diols in order to prevent activation of both ends of the PEG. Likewise, Lee does not disclose purifying monofunctionally activated PEG prior to coupling it to a peptide, protein or glycoprotein. In view of the disclosures in Lee, one of ordinary skill in the art would have understood from Lee that *less than* 95% of the PEG diol would have been activated at only one end. In fact, the binomial statistics of end-group activation prevent more than 50% of the PEG diol from becoming monofunctionally activated in an activation reaction as disclosed in Lee. In the method disclosed in Lee, when the maximum of 50% monoactivation is reached, 25% of the PEG diols would be *bis*-activated, and 25% of the PEG diols would remain unactivated. As one increases the extent of activation further, the fraction of PEG diols that is *bis*-activated would increase to more than 25%, while the fraction of monoactivated PEG would *decrease*, as would the fraction

of unactivated PEG. This is illustrated quantitatively in McManus *et al.*, EP 1 656 410 B1 (hereinafter "McManus," a copy of which is attached as Exhibit B) at page 5 (paragraph [0028]) and page 9 [paragraph [0077]), and in Figure 1. The binomial statistics of diol activation that are illustrated in McManus have long been known.

6. Activating both ends of the PEG diols in Lee would have resulted in the intramolecular or intermolecular cross-linking of PEG to ovalbumin or to ragweed antigen. That is, *both ends* of at least a portion of the activated PEG would have bound covalently to one or two molecules of ovalbumin or to one or two molecules of ragweed antigen, respectively.

7. The PEG molecules utilized in Example 3 of Lee (starting at column 15, line 1) are methoxypoly(ethylene glycols) or mPEGs, the single terminal hydroxyl group of which was activated to permit coupling to dog serum albumin. Such conjugates would have methoxyl groups, not hydroxyl groups, at the distal termini of the coupled PEG.

8. For at least these reasons, Lee fails to teach a composition in which: (i) at least 95% of the polyalkylene glycol(s) is (or are) attached to the peptide, protein or glycoprotein at a single site on the polyalkylene glycol(s), and (ii) a hydroxyl group is present on at least 95% of the distal polyalkylene glycol termini in the conjugate.

9. At page 4 of the Office Action, the Examiner cited Pepinsky *et al.*, WO 00/23114 (hereinafter "Pepinsky"). At the Examiner interview on October 19, 2010, which I attended, the Examiner discussed his basis for the rejection over Pepinsky. It is my understanding that the Examiner interprets Pepinsky's statement that the polymer "may have at least one terminal hydroxyl group" to refer to the status of the polymer *after* activation of the polymer.

10. Under the Examiner's interpretation of Pepinsky, after the polymer is activated, the polymer contains at least one terminal hydroxyl group, and after the activated polymer is coupled to interferon-*beta*-1a, the polymer still contains at least one terminal hydroxyl group. I respectfully disagree with the Examiner's interpretation, as it applies to polyalkylene glycols. Pepinsky does not disclose the polymer recited in the present claims. As the Examiner acknowledged during the interview, Pepinsky's disclosure of "polymers" is quite broad, since Pepinsky included polyols, such as dextrans and other carbohydrates, among the polymers said to be suitable for coupling to interferon-*beta*-1a. Since polyols, such as dextrans and other carbohydrates, can have many hydroxyl groups (some of which may be terminal hydroxyl groups), Pepinsky's use of the term "polymers" cannot properly be interpreted as synonymous with the polyalkylene glycols used to make the compositions of the present invention, in which a hydroxyl group is present on at least 95% of the distal polyalkylene glycol termini in the conjugate in the composition.

11. One of ordinary skill in the art would have understood from Pepinsky at page 18, lines 17-21 that, even if the polymer discussed at page 19, line 13 of Pepinsky has a single terminal hydroxyl group, it is that terminal hydroxyl group on the polymer that is "activated." As a result, the activated polymer discussed at page 19 of Pepinsky could not have contained both a single group that is reactive with proteins *and* a terminal hydroxyl group following activation of the polymer. Similarly, the polymer could not have contained a terminal hydroxyl group after the polymer was attached to a single interferon-*beta*-1a at a single site on the polymer, since one of Pepinsky's polymers that has multiple hydroxyl groups would have been made reactive at many hydroxyl groups, if not at every hydroxyl group.

12. At page 6 of the Office Action, the Examiner cited Delgado *et al.*, U.S. Patent No. 5,349,052 and Zalipsky, which I understand the Examiner to have later confirmed orally to our attorney, Dr. Grant Reed, to be Zalipsky *et al.*, *Eur. Polym. J. 19*: 1177-1183 (1983) (hereinafter "Zalipsky").

13. Delgado *et al.* relates to the activation and coupling of mPEG to GM-CSF, consequently producing a conjugate in which the mPEG has a methoxyl group, not a hydroxyl group, at the distal terminus of the coupled PEG. Therefore, Delgado fails to disclose a composition in which a hydroxyl group is present on at least 95% of the distal polyalkylene glycol termini in the conjugate. Delgado also fails to disclose that the conjugate in such a composition would have exhibited *reduced antigenicity*, compared to a conjugate comprising the same peptide, protein or glycoprotein linked at the same site or sites on the peptide, protein or glycoprotein to the same number of polyalkylene glycols of the same size and the same linear or branched structure, in which a hydroxyl group is present on less than 95% of the distal polyalkylene glycol termini in the conjugate.

14. Zalipsky relates to the attachment of certain small molecule drugs (not peptides, proteins or glycoproteins) to certain PEG molecules. Those PEG molecules are either (a) a PEG with two hydroxyl end groups, *i.e.*, a PEG diol, or (b) a PEG with one methoxyl end group and one hydroxyl end group, *i.e.*, a monomethoxyPEG (also known as "mPEG," which is a type of alkoxyPEG). *See* Zalipsky at page 1171, left column, last paragraph to page 1171, right column, first paragraph.

15. When a PEG diol was coupled to a small molecule drug in Zalipsky, both ends of the diol would have been activated, with the result that a hydroxyl group is *not* present on at least 95% of the distal polyalkylene glycol termini in the conjugate.

Therefore, Zalipsky would not have suggested making a conjugate in which a hydroxyl group is present on at least 95% of the distal polyalkylene glycol termini in the conjugate.

16. Activating both ends of a linear PEG diol would have resulted in the cross-linking of two molecules of the drug by PEG. That is, *both ends* of the activated PEG would have been bound covalently to the drug, forming what is called a "dumbbell" structure (Drug-PEG-Drug). Therefore, Zalipsky would not have suggested making a conjugate in which at least 95% of said polyalkylene glycol(s) is (or are) attached to said peptide, protein or glycoprotein at a single site on said polyalkylene glycol(s).

17. In fact, Zalipsky discloses that a free hydroxyl ("OH") group was *not* present, because Zalipsky reported "no absorption for OH at 3300-3500 cm^{-1}." *See* Zalipsky at page 1177, right column, second full paragraph; page 1178, right column, second full paragraph.

18. When a monomethoxyPEG was coupled to a small molecule drug in Zalipsky, the hydroxyl end of the monoalkoxyPEG would have been activated and coupled to the drug, leaving the alkoxyl moiety in the resultant alkoxyPEG conjugate at the distal terminus of the PEG. A hydroxyl group would not have been present on at least 95% of the distal polyalkylene glycol termini in such a conjugate.

19. Zalipsky's data also indicate that cross-linking occurred, which is distinct from and not suggestive of the presently claimed composition, since Zalipsky reported that the molar ratio of drug to PEG was twice as high in the conjugates prepared using a PEG diol as the starting material compared with conjugates prepared using mPEG as the starting material (*i.e.*, prior to the activation of the polymer). *See* Zalipsky at

page 1178, right column, first paragraph; page 1178, right column, second full paragraph; page 1178, right column, third to last paragraph; page 1179, left column, first full paragraph; and page 1179, left column, second full paragraph.

20. Finally, Zalipsky fails to disclose a PEG-drug conjugate that exhibits reduced antigenicity, compared to a conjugate comprising the same drug linked at the same site or sites on the drug to the same number of polyalkylene glycols of the same size and the same linear or branched structure, in which a hydroxyl group is present on less than 95% of the distal polyalkylene glycol termini in said conjugate.

21. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the present patent application or any patent issued thereon.

Respectfully submitted,

Date: March 3, 2011 Mark Saifer

Mark G.P. Saifer, Ph.D.

Exhibits:

Curriculum Vitae for Mark G.P. Saifer, Ph.D.
McManus *et al.*, EP 1 656 410 B1

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